adenosine derivative with light results in formation of an excited-state compound that can undergo a variety of reactions within the time scale of its fluorescent lifetime, including deprotonation, rotational diffusion, and energy transfer to paramagnetic and colored metal cations near the chromophore. The rates of these reactions are a function of the excited-state lifetime and environmental factors such as the distance between the interacting molecules and the diffusional properties of the bound species. A comparison of the rates of the excited state decay when bound to the enzymatic site to those in solution reveals information of the binding site with respect to the spacial organization (distances between the metal and nucleotide) and the temporal organization (how fast the bound molecules diffuse).

In solution, high concentrations (>50 mM) of Ni(II), Co(II), and Mn(II) cations quench the fluorescence of ethenoadenosine derivatives by collisional quenching. At low concentrations (10^{-6}–10^{-3} M) quenching occurs when the metal binds to the phosphates. The affinity constant depends upon the metal ion and the pH. The increase in the decay rate of the excited-state molecule by the bound metal is primarily a function of the spectral overlap in the metal absorption and the ethenoadenosine fluorescence emission. The effect on intersystem crossing rates by the unpaired electrons of the metal appears to be less significant.

The pK of ethenoadenosine in the excited state is lower than in the ground state. Excitation of the acid form results in emission from the base form, indicating that deprotonation occurs rapidly. Measuring fluorescence decay rates of the ethenoadenosine derivative as a function of pH allows one to calculate the rate of protonation and deprotonation of the excited-state species and then to compare with the quenching rates of the metal cations. The collisional rates for quenching are of the order $\text{Ni} \simeq \text{Co} > \text{H}^+ > \text{Mn}$. It is interesting to note that the pH dependence of ethenoadenosine, etheno AMP, etheno ADP, and etheno ATP differ, indicating that the conformation of the molecule allows interaction between the adenosine ring and the phosphate.

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RUPTURE DIAPHRAGMLESS APPARATUS FOR PRESSURE-JUMP RELAXATION MEASUREMENT

Tatsuya Yasunaga and Nobuhide Tatsumoto, *Department of Chemistry, Faculty of Science, Hiroshima University, Hiroshima, Japan 730.*

The kinetics of micelle formation and dissociation in solutions of anionic surfactants, such as sodium dodecyl and tetradecyl sulfates (SDS and STS), have been studied experimentally by various techniques. Recently a new theory of micelle formation mechanism was proposed together with experimental results obtained in the course of chemical relaxation studies of micellar solutions of ionic surfactants by Aniansson

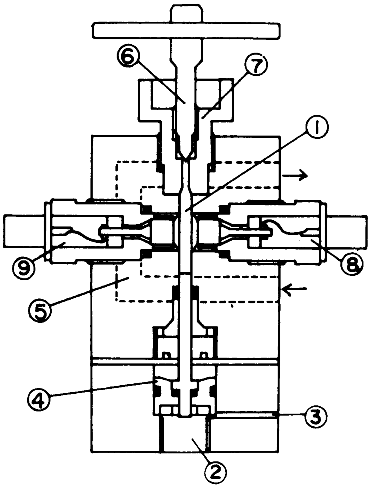

FIGURE 1 1, experimental chamber; 2, gas pressure inlet; 3, gas pressure outlet; 4, piston; 5, heat exchanger; 6, bubble remover; 7, socket; 8, 9, conductivity cells.

et al. (1). In their study the validity of the theory was maintained from the agreement between theoretical and experimental values. It seems to be a successful interpretation of their data. A large discrepancy, however, was found between their experimental data and ours, obtained from the surfactant synthesized extremely purely; i.e. their reciprocal relaxation time (τ_2^{-1}) is dependent upon the concentration of surfactant, whereas ours is independent of the concentration. This fact demanded the reexamination of their theory. Here, the value of τ_2^{-1} is vague near the concentration C^* for SDS and STS because the relaxation strength decreases. The purpose of this study is to develop a new pressure-jump apparatus capable of measuring the relaxation time for small relaxation strengths and to reconfirm the micellar relaxation mechanism by using the newly obtained data.

In the usual pressure-jump method, a single rectangular step-forcing function is realized by blowing out a rupture diaphragm. The decay time of pressure of this method is less than 100 μs, but the precise measurement of relaxation time is impossible in the case of a relaxation of a poor signal-to-noise (S/N). The repetitive application of pressure perturbation and signal averaging is used to enhance the S/N of relaxation curve. The apparatus was designed to produce 100–200 atm of water in an autoclave with a piston (surface area ratio 1:25) by 5–9 atm of air. A cross-sectional diagram of the autoclave is given in Fig. 1. To obtain a short rise time, an electric solenoid valve and gas reservoir were set close to a gas inlet 2. Air in the gas inlet 2 always went out an outlet, 3, but the pressure in the chamber 1 was kept constant as long as the valve was opened, since the air supply was much greater than the exhaust. The signal detected by an AC bridge was rectified by a ring demodulator[1] and averaged with a

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[1] Tatsumoto, N., K. Takehara, and T. Yasunaga. In preparation.

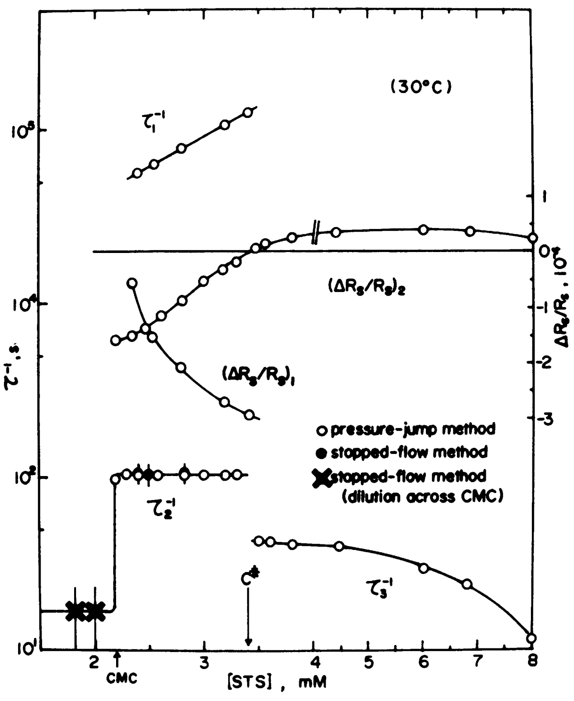

FIGURE 2

signal memory averager. The rise and decay times of pressure in the autoclave were determined to be within 5 and 40 ms, respectively, with use of $MgSO_4$ and NaCl solutions. Although this rise time is longer than that of the usual method, the new technique has the outstanding merit that the repetitive measurement can be performed in a short time. In this experiment, STS was studied. Since τ_2 is about 10 ms at 30°C, the pressure pulse-width was set at 100 ms. The repeating total time is about 25 s for signal averaging of 128 times. The relaxation strength $\Delta R_s/R_s$ and its characteristics were also measured with a usual pressure-jump system.

The relaxation effect characterized by τ_2 vanished around the concentration of C^*. With further increase of the concentration, another new relaxation effect characterized by τ_3 appeared. This phenomenon was also observed in the relaxation strength; i.e., the sign of $\Delta R_s/R_s$ changed from negative to positive with increase of the STS concentration at C^*, as shown in Fig. 2. τ_2^{-1} is clearly independent of and τ_3^{-1} decreases with the concentration. The electroconductometric stopped-flow measurement yielded the same result for τ_2 as that in the pressure-jump method above the critical micelle concentration (CMC) and new relaxation effect, designated by (X) in Fig. 2, was also observed in diluting across the CMC.[2] Taking account of these new

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[2] Tatsumoto, N., T. Tasunaga, and T. Nagamura. In preparation.

experimental results, the mechanism of the micelle formation of surfactants will be discussed.

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